IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re patent application of inventor Saraf

Serial No. 09/870,986 Group Art Unit: 1634

Filed: 06/01/2001 Examiner: Chakrabarti

For: ***"BIO-CHIP, PHOTOLUMINESCENT METHODS FOR IDENTIFYING BIOLOGICAL MATERIAL, AND APPARATUS FOR USE WITH SUCH METHODS AND BIO-CHIPS"***

Assistant Commissioner for Patents
Washington, D.C. 20231

AMENDMENT UNDER 37 C.F.R. § 1.111

Dear Sir:

In response to the Office Action mailed on 02/28/2002, please amend the above-identified patent application as follows:

IN THE CLAIMS:

Please amend the following claims: 1 and. A clean copy of the amended claims is attached.

1. (Amended). A tagging-free method to detect binding of molecules, comprising the steps of:

(A) providing a sensor comprised of a first layer [including a single stranded nucleic acid sequence] and a second layer [including a photoluminescent material] wherein said first layer comprises a single stranded nucleic acid sequence and wherein said second layer comprises a photoluminescent material;

(B) exposing said sensor to a biological sample for sufficient time for said single stranded nucleic acid sequence to bind to a material of interest in said biological sample;

(C) [exposing said sensor to] applying light to said sensor; and

(D) measuring photoluminescence from said sensor, wherein photoluminescence

measured in said step of exposing is indicative of binding of molecules.

3 (Amended). The tagging-free method of Claim 1 wherein said second layer [is] comprises material selected from the group consisting of aromatic polymers, doped or undoped metal oxides, sulfides, selenides, arsenides, tellurides, and nitride and phosphide nanocomposites.

11 (Amended). The tagging-free method of claim 1, wherein said [measuring step includes sensing photoluminescent light from the second layer when] light is applied to said first layer of said sensor, and said light is ultraviolet light with wavelength in the range of 200-700nm [is applied to the first layer].

12 (Amended). The tagging-free method of claim 11, wherein the wavelength of the ultraviolet light [applied] is in the range of 260-265 nm.

20 (Amended). The tagging-free method of claim 1, [including providing a discontinuous first layer comprising different nucleic acid sequences in different sections of] wherein said first layer comprises a plurality of sections each of which comprises a different nucleic acid sequence.

REMARKS

The application includes claims 1-23, which represent Applicant's previous election, without traverse, of the "Group I" claims.

35 U.S.C. § 112, second paragraph rejection

Claims 1-23 stand rejected under 35 U.S.C. § 112, second paragraph, as being indefinite for failing to particularly pont out and distinctly claim the subject matter which applicant regards as the invention. In particular, claim 1 has been rejected as indefinite because, according to the Examiner, it lacks a final process step that clearly relates back to the preamble.

Claim 1 has hereby been amended to recite that the photoluminescence measured in the

(new, see below) step of measuring is indicative of binding of molecules as set forth in the preamble. Applicant submits that the inclusion of this phrase in the last step of the claim clearly states that the goal of the claim as stated in the preamble is accomplished by the method, thereby overcoming Examiner's rejection.

Claims 1, 11 and 20 stand rejected over the recitation of the terms "including" and "includes". Examiner states that the use of these terms renders the claims indefinite because it is unclear whether the limitation(s) following those terms are part of the claimed invention.

Claims 1 has hereby been amended to eliminate the word "including" and now recites that the sensor comprises a first and an second layer, and then additional characteristics of the two layers are particularly spelled out by reciting "wherein first layer..." and "wherein second layer..." comprises a single stranded nucleic acid sequence and a photoluminescent material, respectively. Applicant submits that this amendment renders the meaning of the claimed invention clear, and that any confusion regarding the relationship of those particular features to the claimed invention has been eliminated.

Claim 1 has further been amended to provide additional clarification. The previous step of "exposing said sensor to light and measuring photluminescence from said sensor" has been broken into two steps: 1) applying light to said sensor (the word "applying" has been substituted for exposing in order to distinguish this step from the previous step of exposing; Applicant submits that the application of light has precedent in, for example, original claim 11 where light is described as "applied", and does not therefore constitute new matter); and 2) measuring photoluminescence. Applicant submits that this second step is merely the removal of the step of measuring from within the previous step of "exposing said sensor to light", and therefore also does not constitute the introduction of new matter.

Claim 11 has hereby been amended to omit the phrase "measuring step includes sensing photoluminescent light from the second layer" and rather to recite simply that the "light" (from step (C) of claim 1 may be applied to the first layer, and to specify that the light may be ultraviolet of the indicated wavelength range. Applicant submits that this simplification does not add any new material and thus does not constitute new matter, and serves to simplify and make

more direct the meaning of the claim, thereby overcoming Examiner's rejections.

Claim 20 has hereby been amended to omit the phrase "including providing...". Claim 20 now recites the same limitation (the first layer of the sensor may contain different nucleic acid sequences arranged in different sections of the layer) but in language which is more direct. The amended claim now recites that the first section may contain "a plurality of sections" (i.e. more than one section) and that each section may contain a different nucleic acid sequence. Applicant submits that the "meaning" and scope of the claim have not been altered, but that the arrangement of words has been changed to increase clarity. Thus, this amendment does not constitute that introduction of new matter. Applicant submits that this amendment adequately addresses Examiner's rejection.

Claim 8 stands rejected due to the recitation of the phrase "group II and group VI". Examiner states that this reference is unclear, given the absence of any "groups" in the specification. Applicant respectfully disagrees. The phrase "group II and group VI" is found in the sentence at lines 8-10 on page 6, and reads: "In a further preferred embodiment, the photoluminescent particles may be doped or undoped compounds selected from the group consisting of group II and group VI." Applicant submits that, by clearly referring to the materials in question as being "photoluminescent particles" which are either doped or undoped compounds, one of ordinary skill in the art would immediately recognize that a reference to a "group" further denominated by a Roman numeral would be referring to a group of the Periodic Table of the Elements. Applicant further submits that a person of ordinary skill in the arts relevant to the instant invention would of course, be familiar, with the Periodic Table. In fact, to those with even a rudimentary (e.g. freshman college level) background in chemistry, the phrase "group II" or "group VI" when referring to "compounds" can refer only to the groupings of elements found in the Periodic Table. Further, specific examples of elements in those groups are recited in, for example, claim 3 and in the specification at page 5, lines 30-33 (selenides and tellurides, group VI) and on page 6, line 11 (zinc, group II). Thus, Applicant submits that the meaning of the phrase "group II and group VI"is not "vague and indefinite" as suggested by Examiner.

In view of the foregoing, reconsideration and withdrawal of this rejection are respectfully requested.

Other amendments:

Claim 3 has been amended to eliminate the word "is" from the phrase "said second layer is selected from ..."and to recite "said second layer *comprises* material selected from...". This change has been made in order to more accurately describe the invention. The second layer of the sensor of the present invention may be comprised of more than one material (see, for example claim 7 where the second layer *comprises* photoluminescent particles in a polymer matrix). Further, on page 5, last paragraph, and continuing on to the first paragraph of page 6, various materials which may be used in the fabrication of the second layer are described, and clearly some may be combined (e.g. a "photoluminescent material associated with said matrix material", page 6, line 2). Thus, the use of the term "is" in claim 3 is not accurate since the listed materials would not be the sole component of the second layer. Rather, the second layer may include (i.e. "comprise" those materials, in combination with, e.g. a photoluminescent material.

Applicant respectfully requests entry of this amendment.

35 U.S.C. §103(a) rejections (Examiner's points 5-7)

Examiner's point 5. Claims 1-5, 7, 13-17 and 19-20 stand rejected under U.S.C. 35 §103 over Leland et al. (US Patent 5,962,218) in view of Charra (US Patent 5,831,259). Examiner states that Leland discloses a method to detect binding of molecules with the steps of providing a sensor comprised of single stranded nucleic acid sequences and a photoluminescent material, exposing the sensor to a biological sample, and exposing the sensor to light and measuring photoluminescence from the sensor. Examiner further states that Leland et al. do not teach a tagging-free method. Examiner asserts that Charra teaches a tagging free method, providing a first layer of oligomer and a second layer of photoluminescent material which consists of aromatic polymers embedded in the matrix material. Examiner further states that it would have been obvious to combine the teachings of Leland et al. with those of Charra, and that

such a combination would result in the present invention.

Applicant respectfully disagrees.

The subject matter of the claims which have been rejected under Examiner's Point #5 is a tagging-free method to detect the binding of molecules. By "tagging-free" method it is meant that no extrinsic tagging or labeling of the molecules which undergo binding is necessary in order to detect bound molecules. Rather, changes in the intrinsic properties of bound vs unbound states of the molecules are taken advantage of. The intrinsic properties with respect to DNA are discussed in detail on page 12, lines 1-32 of the speicifcation. Briefly, Applicants have discovered that upon conversion of ssDNA to dsDNA by the binding of a complementary ssDNA strand, not only do the light absorption properties of the DNA change (as is well known) but also the refractive index, (and thus reflectivity and scattering) of radiation changes to an even greater extent. Thus, the amount of light reflected away from ssDNA vs dsDNA is dramatically different (the change may be as high as 200%) so that any material adjacent to the DNA will receive a correspondingly different amount of light reflected from the DNA upon the ss to ds conversion. In order to capitalize on this observation, the present invention provides a photoluminescent layer to which a layer containing the ssDNA is attached. While the DNA remains single stranded, light which impinges on the ssDNA will be reflected onto the photoluminescent layer at a certain level, causing a particular degree of luminescence from the layer. Upon conversion of the ssDNA to dsDNA, the same amount of impinging light on the dsDNA will result in a dramatically different level of light reflected from the dsDNA and onto the photoluminescent layer, causing a change in the degree of luminescence from the layer. The change in luminescence from the photoluminescent layer can be detected before and after exposure of the two layer (ssDNA and photoluminescent) sensor to a sample of interest. A change in luminescence is thus indicative of the occurrence of a binding event, for example, a change in the state of DNA from ssDNA to dsDNA. By this method, a binding event may be detected without the necessity of providing any external labeling moiety as is required in the binding detection methods known prior to the instant invention.

One such previously known method is that of Leland et al., which has an absolute

requirement for an extrinsic label. In particular, each claim of Leland et al. requires, and each example shows the use of, an electrochemiluminescent (ECL) label.

Examiner states that Leland discloses a method to detect binding of molecules with the steps of providing a sensor comprised of single stranded nucleic acid sequences and a photoluminescent material, followed by two steps of exposing, apparently in order to equate the method of Leland et al. to that of claim 1 of the instant invention. Applicant respectfully submits that Examiner has not taken into account the precise wording of claim 1, which states that the sensor comprises a first layer which includes ssDNA and a second layer which includes a photoluminescent material. Applicant submits that corresponding elements are not disclosed by Leland et al., and that the method of Leland et al. differs markedly from the instant invention.

Three specific prototypic embodiments of Leland et al. are presented in column 19, lines 30-45, and all the ensuing examples are based on these:

I. The first is described with reference to Figure 3, and has the following steps:

1) dsDNA products from a PCR reaction are labeled with both biotin and an ECL label;

2) the dsDNA is then "captured" (via the biotin moiety) by streptavidin beads;

3) the beads are washed;

4) the ECL label is detected.

II. The second embodiment is described with reference to Figure 4, and has the following steps:

1) a PCR product is labeled with biotin;

2) the labeled product is captured by streptavidin beads;

3) the strand without biotin is removed;

4) the remaining bead-bound ssDNA hybridizes with a complementary ssDNA that is labeled with ECL;

5) ECL label is detected.

III. The third embodiment is described with reference to Figure 5, and has the following steps:

1) ssDNA is labeled with biotin, and a complementary ssDNA is labeled with ECL;

2) the two complementary strands are allowed to hybridize;

3) the hybridized dsDNA is captured by streptavidin beads;

4) ECL label is detected.

A schematic depiction of these embodiments, of which all other examples in Leland et al. merely constitute "real life" experiments, is included herewith as Appendix I for Examiner's convenience. Claim 1 of the present invention requires a first layer which comprises a ss nucleic acid. As can be seen from the above analysis, this feature is not present in any embodiment of Leland et al. The ssDNA in the various steps of the method of Lelande et al. is, in the first embodiment, free in solution and labeled with biotin or ECL (or both); in the second embodiment, it is attached to a streptavidin bead prior to hybridization. It is never a component of a "layer" of a sensor. Claim 1 requires a second layer which comprises a photoluminescent material. This feature is also not present in any embodiment of Leland et al. The only "photoluminescent material" in Leland et al. is the ECL label. In the embodiments depicted by Leland et al., the ECL is either attached directly to a ssDNA or a dsDNA molecule that is free in solution, until the final complex as been formed. The ECL is never a component of a second "layer" of a sensor. Leland et al. does not show or suggest the equivalent of the first ssDNA containing layer and the second photoluminescent material containing layer of the present invention.

Applicant is aware that it is the combination of Leland et al. with Charra which forms that basis of Examiner's rejection. Examiner asserts that Charra supplies the tagging-free aspect of the present invention, by providing a "first layer of oligomer and a second layer of photoluminescent material consisting of aromatic polymers embedded in the matrix material".

Applicant strongly disagrees with Examiner's description of the invention of Charra. What Charra describes is a high impedance electrooptical transducer that includes an electrosensitive element which emits photoluminescence when illuminated by excitation radiation. Photoluminescent conjugate oligomers intermediate in size between those of conventional small molecule fluorophores and the much larger semiconductor-type polymers are employed. With this size of oligomer, the creation of a state resulting in fluorescence and the creation of a state which is only slightly fluorescent are about equally probable, and the probabilities of the occurrence of each is highly dependent on the presence of an electric field.

Thus, the generation of an electric field in the presence of a film of such polymers causes a variation in the photoluminescence efficiency of the oligomers in the film. The detection of such variations is useful in many systems. Examiner states that Charra describes a "first layer of oligomer and a second layer of photoluminescent material consisting of aromatic polymers embedded in the matrix material". This is incorrect. Charra describes an electrosensitive element comprising "a layer containing said oligomers" (column 3, lines 8-9). The oligomers themselves are "photoluminescent conjugate oligomers" (column 2, line 25-26) which are described in detail in column 2, lines 62-65. Thus, the "oligomers" and the "photoluminescent material consisting of aromatic polymers" described as two separate elements by Examiner are one and the same. There is not a separate first layer of "oligomers" and a separate second layer of other "photoluminescent material consisting of aromatic polymers ". There is a single layer or film formed from "photoluminescent conjugate oligomers". In the figures referred to by Examiner, (and in all figures of the patent, this film is represented by the numeric indicator 4, and there is no second layer answering to either of Examiner's descriptions. Other features of the figures which might appear to be "layers" (e.g. in Figure 1) are 6, 8, and 10, which are in fact electrodes; and 2, which is a piezoelectric shim (see description in column 6, lines 19-25, for example). In short, there is no "first layer of oligomer and a second layer of photoluminescent material consisting of aromatic polymers embedded in the matrix material" disclosed in Charra. Further, there is no showing or suggestion in Charra that the film that is described could be useful for purposes of developing a method to detect the binding of molecules of any type.

Applicant submits that there is no meaningful way to combine Leland et al and Charra. There is no place in the invention of Leland et al. for the inclusion of a film of photoluminescent conjugate oligomers. Which element(s) of Leland et al. could be replaced by or made to include the film? The ECL label? The polystyrene bead? The DNA? The modification proposed by the Examiner would destroy the function or operation of the two references.

Further, Applicant submits that Examiner's suggestion that one of skill in the art would be motivated to combine Leland et al. and Charra in order to achieve the "express advantages ... of an invention that provides use of reduced size oligomers which decreases the mobility of the

charges and therefore minimizes energy consumption" is totally without foundation. Minimize energy consumption of what? To Applicant's knowledge, the minimization of energy consumption in assay methods such as those of the instant invention (e.g. biochips) is not a consideration. There is no motivation for decreasing the energy required for the hybridization of ssDNA, or for the reflection of light from the hybridized dsDNA to a juxtaposed layer of photoluminescent material, which processes form the heart of the instant invention.

In sum, neither Leland et al. or Charra either show or suggest a tagging free method to detect the binding of molecules which includes providing a sensor comprising two layers, one which includes ssDNA and the other of which includes a photoluminescent material. Thus, the combination of these two references cannot render the present invention obvious.

In view of the foregoing, reconsideration and withdrawal of this rejection are respectfully requested.

6. Claims 1-7, 11-17 and 19-20 stand rejected over Leland et al in view of Charra and further in view of Leising et al.

Examiner's comments concerning Leland et al. and Charra and Applicant's response is given above under Point #5. Examiner states that Leising et al. further supplies the teaching of a matrix layer comprising polystyrene. Applicant concurs. Leising et al. do teach a matrix layer comprising polystyrene. However, for the reason outlined above, this point is moot. Applicant submits that there is no showing or suggestion of a "matrix layer" or equivalent element in either Leland et al. or Charra, and no way to combine those two references to achieve such a layer, that could then be improved by being fashioned from polystyrene.

In view of the foregoing, reconsideration and withdrawal of this rejection are respectfully requested.

7. Claims 1-5 and 7-23 stand rejected over Leland et al. in view of Charra and further in view of Bhargava et al.

Examiner's comments concerning Leland et al. and Charra and Applicant's response is

given above under Point #5. Examiner states that Bhargava et al. further supplies the teaching of utilizing doped or undoped zinc sulfide, and the use of ultraviolet light with a "wavelength in the range of 200-700nm". (Applicant notes that such a wavelength range includes much more than ultraviolet light.) Again, Applicant does not dispute that Bhargava et al. teach these elements. However, as discussed above under Point #5, there is no reasonable combination of Leland et al. and Charra with which to combine Bhargava et al., thus the teaching of Bhargava et al. is moot.

In view of the foregoing, reconsideration and withdrawal of this rejection are respectfully requested.

Formal Matters and Conclusion

In view of the foregoing, Applicant submits that all rejections have been successfully traversed. The Examiner is respectfully requested to pass the above application to issue at the earliest possible time.

Should the Examiner find the application to be other than in condition for allowance, the Examiner is requested to contact the undersigned at the local telephone number listed below to discuss any other changes deemed necessary in a telephonic or personal interview.

Please charge any underpayment or credit any overpayment of fees to attorney's deposit account #50-2041.

Respectfully submitted,

Ruth E. Tyler-Cross

Reg. No. 45,922

Whitham, Curtis & Christofferson

30743

PATENT TRADEMARK OFFICE